FISSION YEAST MODEL RECAPITULATES ASPECTS OF α-SYNUCLEIN MISFOLDING

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Budding yeast has emerged as an excellent model to probe protein misfolding linked to neurodegenerative diseases, including Huntington's disease, prion diseases, and recently, Parkinson's disease (PD). In contrast, fission yeast models have not been reported for any neurodegenerative disease. Since α-synuclein misfolding and aggregation is linked to PD, this project developed an *Schizosaccharomyces pombe* model to evaluate α-synuclein misfolding. We expressed wild-type (WT) α-synuclein and three mutants (A53T, A30P, A53T/A30P) using the thiamine repressible NMT1 promoter (and its weaker variants, NMT41 and NMT81) to test the hypothesis that α-synuclein misfolds and aggregates *in vivo* in a concentration dependent fashion. As expected, higher amount of α-synuclein was expressed with increased promoter strength. When driven by NMT1 promoter, GFP-tagged WT and A53T α-synuclein accumulated as ~1-5 foci per cell, evidenced as *in vivo* aggregation, whereas A30P and A53T/A30P were diffusely cytoplasmic. The number of these foci-positive cells increased over time and, compared to wildtype α-synuclein, A53T accumulated foci earlier in the time course. Repression of α-synuclein expression, achieved either by exposing NMT1 driven cells with increasing thiamine or using NMT41 or NMT81 promoters, resulted in dose-dependent decrease in number of cells with foci. Surprisingly, α-synuclein expression was not significantly toxic to this yeast. As confirmed in several wildtype *pombe* strains, these live yeast studies support a nucleation-polymerization model of aggregation for α-synuclein. Surprisingly, unlike our *S cerevisiae* findings (Herrera *et al.*, this meeting), none of the α-synuclein forms localized to the cell membrane in *S pombe*. The two yeasts reveal distinct aspects of α-synuclein biology and are useful for studying molecular regulation for misfolding linked to neurodegeneration. (NSF 0115919 and NIH NS48508)